Vitamin C can reduce toxic effects of Nano Zinc Oxide

Bakhshiani Somayeh1* and Fazilati Mohammad2
1Biology Department, Faculty of Sciences, Payam Noor University of Isfahan, Isfahan, IRAN
2Department of Biochemistry, Payam Noor University of Isfahan, IRAN

Available online at: www.isca.in, www.isca.me
Received 29th October 2013, revised 24th November 2013, accepted 19th January 2014

Abstract

Nanoparticles are extensively employed in most industries and biological sciences. Zinc nanoparticles have widespread application in industries manufacturing medical equipment as well as household because of its unique features such as immediate effect, greater stability, and antimicrobial properties and special factors. Therefore many peoples are exposure to nanoparticles that maybe have harmful side effects. Detrimental effects of zinc oxide nanoparticles were the objective of different previous studies whit various aspects. Aim of the present research was investigation of the damages of zinc oxide nanoparticles on the liver cells and blood factors in Wistar rats. Also this study assessed the role of vitamin C in the reduction of toxic effects of nanoparticles on the mentioned factors. 36 male rats with approximately 35 days age were divided in to six groups with 6 rats separated. The rats in the experimental groups were administrated by two acute doses of nanoparticles 200 and 400 mg/kg. After 7 days rats blood samples were prepared, then AST, ALT, ALP levels measured and WBC and placket numbered, obtained results demonstrated a significant increasing in the number of WBC in the experimental groups compared to the control. Results showed rise of the liver enzymes concentration in the blood but application of vitamin C inhibit this result. This response was dose dependent and was more significant in high dose (p<0.05). Our results demonstrated that zinc oxide nanoparticles interrupt the function of liver cell membrane so cause diffusion of liver enzymes to blood. By using of an antioxidant agent such as vitamin C toxic effects of nanoparticles reduced. This indicates one of more important ways of nanoparticles damaging is oxidative stress.

Keywords: Zinc oxide nanoparticle, vitamin C, liver enzymes, blood factors, wistar rat

Introduction

Nanoparticles have special physical and chemical properties and unusual shape, size and surface area to volume ratio. Mentioned features make these materials unique for biological, medical and industrial applications. One of the most useful features is high surface area that causes nanoparticle’s widespread application in medical sciences and production of nano based drugs for some of the Incurable disease1.

Not that some of nanoparticles such as nano-ZnO have growing production for applicationin biological systems but there aren’t widespread and sufficient studies on the side effects of nanoparticles on the living systems2. We need an appropriate model for assessing of nanoparticles risk3.

Published literatures showed material at the nano size has relatively greater toxicity rather than large sizes materials, because nanoparticles are highly reactive and cause oxidative stress in human and animals. Oxidative stress can cause serious damages in DNA and protein structures there for cause mutation4. Nanoparticles can pass through the cell membrane easily and even pass through blood-brain barrier and blood-testes barrier5,6. They can across the protective layers of the body and enter the circulation7,8 and reach to different organs of the body9. Recent studies showed exposing of rats by 5 g/kg dose of zinc oxide nanoparticle create symptoms such as lethargy, diarrhea, vomiting and even death10. One of the important ways that nanoparticles can damage to the body is production of free radicals or ORS11.

Vitamin C is a water soluble antioxidant agent that accumulates in the brains of mammals more than any other tissue. Babies' brain has its accumulation more than adults12. There are many reports about the reduction of DNA damage probability and preventing of cancer and heart disease by vitamin C because of antioxidant properties13,14.

Liver is central organ in metabolism and detoxification so is one of the organs that damaged in nanoparticles exposing15. Liver damage cause shifting in the level of liver enzymes such as aspartate aminotransferase (AST) and alanine aminotransferase (ALT) so levels of these two enzymes in the blood serum using as an efficient indicator to liver damages and liver diseases16.

In this study we try to presenting a model for action of ZnO nanoparticle in the body especially in the liver by most useful animal model (rat). So we administrated different dose of ZnO nanoparticle to the rats and estimated liver damages by measuring of ALT and AST level. This study also tries to confirm the vitamin C role as an antioxidant agent in reduction of liver damage.

International Research Journal of Biological Sciences ___________________________________
Vol. 3(3), 65-70, March (2014)
Material and Methods

This study is an experimental effort that carried out on animals and we used adult male Wistar rats weighing 300 ± 30 g were estimated from the animal house of martyr portal was developed. Rats whit average age of 2 months selected. Testing carried out at temperature of 22-25 centigrade degree that day duration was 12 hours and 12 hours dark lighting. Municipal tap water was used adjusted drinking water and eating animals for food by rats (feed compression) that the company prepared feed was barking in this study. Rats kept 2 weeks in this condition for adaptation. After this time duration experimental animals were randomly divided into 6 groups with 8 rats in each group as follow: fist control group feed by usual water and food. Second control group that referred to Placebo, injected by 1 ml distilled water very other day intraperitoneally for equivalency of shock that obtained by intraperitoneally injection. Other groups from 3rd and 4th injected by 1 ml ZnO nanoparticles in 200 and 400 mg/kg doses, injection repeated every other day intraperitoneally. 5th and 6th groups received 200 and 400 mg/kg dose of nano-ZnO with 100 mg vitamin C intraperitoneally. Mentioned doses administrated only in one injection acutely. For 5th and 6th groups injection of vitamin C done 1 hour after the nanoparticle injection. Nanoparticles resolved in physiological serum in 20 min by sonicaton method to producing a stable suspension.

7 days after the injection, blood sample of all animals prepared from heart veins (Animals were anesthetized by chloroform before blood sampling). Next the cloting, samples were centrifuged at 3000 rpm for 15 minutes. Subsequently serum separated from the clot by Smplr, serums frozen at temperature of - 20 ° C and stored, then used for measurement. Liver indicator enzymes such as AST and ALT measured by immuno radiometric assay.

The results analyzed based on the statistical program SPSS and analyzed by ANOVA and Tukey test. The differences in the level of P <0.05 was considered significant.

Results and Discussion

For investigation of nano-ZnO effects on the physiological and histological condition of liver, this material injected to the rats intraperitoneally. After administration blood serum of animals extracted and AST, ALP and ALT level of blood measured for estimating of liver condition. The levels of WBC, lymphocyte and placket of blood also measured for create an efficient model for damages of ZnO-nanoparticle in human body.

Result showed ALT level of rats that received 400 mg/kg dose of nano-ZnO significantly increased rather than control group but 6th group that injected by same dose of nanoparticle accompanied by vitamin C with doesn’t show significant enhancement. Therefore application of vitamin C as an antioxidant reagent Inhibits the damaging effects of nanoparticles (figure 1).

Figure 2 showed significant increase in the level of AST enzyme of blood serum in 400 mg/kg does of nano-ZnO. Results showed injection vitamin C decreased nanoparticle effects on the AST level.

Injection of nanoparticle cause significant increase in ALP level of blood in the both of the 200 and 400 mg/kg dose, interestingly injection of vitamin C inhibit nano-ZnO effect to enhancement of ALP level (figure 3).

Between experimental groups, all groups that administrated by ZnO nanoparticle showed improvement of white blood cells number but only rats that received 400 mg/kg does of nanoparticle showed significant increase of WBC number that. This improvement inhibit partially by vitamin C (figure 4).

Injection of nano-ZnO cause significant increase in lymphocyte percentage in the both of the 200 and 400 mg/kg dose, interestingly injection of vitamin C inhibit these enhancements (figure 5).

Figure 6 tell us the groups of rats that injected by 200 and 400 mg/kg does of nano-ZnO showed significant increase in the percentage of placket, this increase in 200 mg/kg dose inhibit by vitamin C but in high dose vitamin C is not affective.

Discussion: Nanoparticles have very specific chemical and physical individualities such as size, shape and high surface area to volume ratio that facilitate its medical and biological applications. Therefore they have widespread and growing application in all aspects of industries and sciences and entering of these materials to environment is inevitable. On the other hand this material distributed in all of body rapidly after injection by circulation and reached to the all of the organs and tissues. So pollution of our living environment by nanoparticles is very dangerous because they can interact with macromolecules in living cells while we don’t have enough information and suitable model for the action of these materials in the body. In this study we tried to give some useful information about their biological actions.

One of the important indicators for assessing of liver health status is presence of alanine aminotransferase, aspartate aminotransferase (both of these are intracellular enzymes in liver cells) and alkaline phosphatase (that is membrane protein in the live cells). Presence of these three enzymes is as a result of damage in liver cells membrane so direct to the unhealthy condition in liver. Nanoparticles as toxic materials can move to the liver and hurt it so AST, ALT and ALP levels in blood increased, our results confirm that. But presence of vitamin C prevents damage to the liver cells. We know vitamin C is an antioxidant agent so results showed damaging effect of ZnO nanoparticle is oxidative destruction. Previous results that agreement with our results confirmed nano-TiO₂ increase the oxidative stress in the cells.
Figure-1
Increasing of ALT enzyme activity in the response to ZnO nanoparticle injection. This enhancement inhibited by vitamin C

Figure-2
Measurement of AST enzyme level in the presence of ZnO nanoparticle showed rise of this enzyme in the 400 mg/kg does
Figure-3
Increasing of ALP enzyme activity in the response to ZnO nanoparticle injection. This improvement inhibited by vitamin C

Figure-4
Increasing of WBC numbering in the 400 mg/kg does of ZnO nanoparticle showed presence of infection in the body
Figure-5
Decreasing of lymphocyte cells in the response to infection that caused by nano-ZnO in the both of acute doses

Figure-6
Reduction of placket cell numbers in the presence of 200 and 400 mg/kg doses of nano-ZnO
Conclusion

Presence of nanoparticles in the body cause involving of immune system and increasing of white blood cells in the blood such occurred in present study; this may be happened because of infection in lymphatic glands. Its interest that application of vitamin C prevents enhancement of WBC so inhibit infection in the body. Our results confirmed anti infection roll of vitamin C in the human body, more of infections are as a result of oxidative agents.

References